

antibody, and the eosinophils and the group of neutrophilic cells are distinguished on the two-dimensional scattergram.--

--13. The method according to claim 1 that in the step (3), a two-dimensional scattergram is produced from the intensity of the scattered light and the intensity of the fluorescence from the first fluorescence-labeled antibody, and the group granulocytic cells are distinguished on the two-dimensional scattergram.--

--14. The method according to claim 1 that in the step (5), a two-dimensional scattergram is produced from the intensity of the fluorescence from the second fluorescence-labeled antibody and the intensity of the fluorescence from the third fluorescence-labeled antibody, and the neutrophilic cells are classified according to the degree of maturity on the two-dimensional scattergram.--

*b2
conceded*

REMARKS

The Examiner reiterated the rejection of claims 1-10 under 35 U.S.C. §102(b) as being anticipated over Bowen, et al and Loken, et al., and also her rejection to claim 11 under 35 U.S.C. §103(a) as being unpatentable over Bowen, et al. and McCarthy, et al.

Applicants have amended claim 1 to place it in better condition for allowance. It respectfully is submitted that the amendment does not introduce new matter and entry and approval of the same is solicited. Support for the amendment can be found in the specification as originally filed at page 13, lines 3-15.

Applicants have also added new claims 12-14. It respectfully is submitted that the new claims do not introduce new matter and entry and approval of the same is solicited. Support

for these new claims can be found in the specification as originally filed at Examples 1 and 2, and accompanying drawing figures.

In view of the foregoing, favorable action on the merits, and allowance of all claims, respectfully is solicited.

Respectfully submitted,

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Inventor: Berend HOUWEN, et al.
U.S. Application Serial No.: 09/388,899
For: METHOD FOR CLASSIFYING AND COUNTING LEUKOCYTES

EXHIBIT 1

"MARKED UP" AMENDMENTS TO CLAIMS PURSUANT TO RULE 1.121(c)



1. (Twice Amended) A method for classifying and counting leukocytes comprising the steps of:

(1) adding to a hematological sample the following fluorescence-labeled antibodies labeled with fluorescent dyes which emit fluorescences distinguishable from each other;

(a) a first fluorescence-labeled antibody which binds specifically to leukocytes,

(b) a second fluorescence-labeled antibody which binds to at least one kind of neutrophilic cells, and

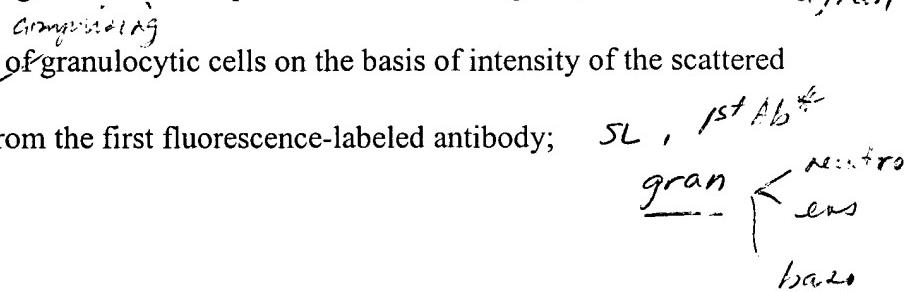
(c) a third fluorescence-labeled antibody which binds to at least one kind of immature granulocytic cells,

in order to stain the leucocytic cells in the hematological sample, and

removing erythrocytes from the hematological sample;

(2) analyzing the resulting hematological sample using a flow cytometer to measure at least one scattered light signal and three separate fluorescence signals;

(3) defining a group of granulocytic cells on the basis of intensity of the scattered light and intensity of fluorescence from the first fluorescence-labeled antibody;



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(4) distinguishing eosinophils and a group of [defining the] neutrophilic cells in the defined group of granulocytic cells on the basis of the intensity of the fluorescence from the first fluorescence-labeled antibody and the intensity of the fluorescence from the second or third fluorescence-labeled antibody; ^{1st Ab * , 2 or 3 Ab *} _{eos, neutro}

(5) classifying (the defined) group of the neutrophilic cells into groups of, ^{1st Ab * , 2 or 3 Ab *} _{neutro}
(neutrophilic cells) different in degree of maturity on the basis of the intensity of the fluorescence from the second fluorescence-labeled antibody and the intensity of the fluorescence from the third fluorescence-labeled antibody, and ^{2nd Ab * , 3rd Ab *} _{"maturity"}
counting the number of cells in each of the groups.

12. The method according to claim 1 that in the step (4), a two-dimensional scattergram is produced from the intensity of the fluorescence from the first fluorescence-labeled antibody and the intensity of the fluorescence from the second or third fluorescence-labeled antibody, and the eosinophils and the group of neutrophilic cells are distinguished on the two-dimensional scattergram.

13. The method according to claim 1 that in the step (3), a two-dimensional scattergram is produced from the intensity of the scattered light and the intensity of the fluorescence from the first fluorescence-labeled antibody, and the group of granulocytic cells are distinguished on the two-dimensional scattergram.

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14. The method according to claim 1 that in the step (5), a two-dimensional scattergram is produced from the intensity of the fluorescence from the second fluorescence-labeled antibody and the intensity of the fluorescence from the third fluorescence-labeled antibody, and the neutrophilic cells are classified according to the degree of maturity on the two-dimensional scattergram.